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#### Henco et al.

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# [54] PROCESS AND DEVICE FOR THE SEPARATION AND DETECTION OF COMPONENTS OF A MIXTURE OF MATERIALS BY TEMPERATURE GRADIENT GEL ELECTROPHORESIS

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#### Related U.S. Application Data

[63] Continuation of Ser. No. 200,551, Feb. 22, 1994, abandoned, which is a continuation of Ser. No. 847,761, filed as PCT/EP90/01366 Aug. 18, 1990, abandoned.

#### [30] Foreign Application Priority Data

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[58]	Field of	Search		43	5/6; 935/77.

#### [56] References Cited

#### U.S. PATENT DOCUMENTS

4,671,870	6/1987	Tompa et al	. 210/149
5,066,377	11/1991	Rosenbaum et al	204/182.8

935/78, 91.2

#### FOREIGN PATENT DOCUMENTS

0198207 3/1986 European Pat. Off. . 0318273 5/1989 European Pat. Off. .

#### OTHER PUBLICATIONS

Val C. Sheffield et al., "Attachment of a 40-base pair G+C-rich sequence (GC-clamp) to genomic DNA fragments by the polymerase chain..." Proc. Natl. Acad. Sci., USA. vol. 86, pp. 232-236, 1989.

Ann-Christine Sylvanen et al., "Quantification of polymerase chain reaction products by affinity-based hybrid collection", Nucleic Acids Research, vol. 16, No. 23, pp. 11327–11338, 1988.

Roger M. Wartell et al.. "Detecting base pair substitutions in DNA fragments by temperature-gradient gel electrophoresis". Nucleic Acids Research, vol. 18, No. 9, pp. 2699–2705, 1990.

Richard M. Myers et al., "Nearly all single base substitutions in DNA fragments joined to a GC-clamp can be detected by . . . ", Nucleic Acids Research, vol. 13, No. 9, pp. 3131-3145, 1985.

Richard M. Myers et al., "Detection & Localization of Single Base Changes by Denaturing Gradient Gel Electrophoresis", Methods in Enzymology, vol. 155, pp. 501–527, 1987.

Detlev Riesner et al., "Temperature-gradient gel electrophoresis of nucleic acids: Analysis of conformational transitions, sequence variations, . . . ", Electrophoresis, vol. 10 (5-6), pp. 377-389, 1989.

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#### [57] ABSTRACT

The invention relates to a process for the separation and detection of components of a mixture of materials by temperature gradient gel electrophoresis, wherein either

- a spatial temperature gradient is built up by spatially separated temperature levels, or
- a time temperature gradient, or
- a temperature gradient is built up by combination of spatial and timewise temperature gradient.

The temperature levels for building up the spatial temperature gradient are adjusted by controllable heating or cooling devices.

To build up the time temperature gradient, the temperature level at each point of the separation path within the separation medium may be optionally adjusted time-dependently by means of controllable heating or cooling devices. There is described a device for performing the process with controllable heating or cooling devices to build up temperature gradients, a hollow body arranged between the temperature levels which contains the medium used for separation, and a thermostat jacket enclosing the hollow body.

12 Claims, 16 Drawing Sheets